

Effects of Calcium and Dairy on Body Composition and Weight Loss in African-American Adults

Michael B. Zemel, Joanna Richards, Anita Milstead, and Peter Campbell

Abstract

ZEMEL, MICHAEL B., JOANNA RICHARDS, ANITA MILSTEAD, AND PETER CAMPBELL. Effects of calcium and dairy on body composition and weight loss in African-American adults. *Obes Res.* 2005;13:??-?.

Objective: Our objective was to determine the effects of dairy consumption on adiposity and body composition in obese African Americans.

Research Methods and Procedures: We performed two randomized trials in obese African-American adults. In the first (weight maintenance), 34 subjects were maintained on a low calcium (500 mg/d)/low dairy (<1 serving/d) or high dairy (1200 mg Ca/d diet including 3 servings of dairy) with no change in energy or macronutrient intake for 24 weeks. In the second trial (weight loss), 29 subjects were similarly randomized to the low or high dairy diets and placed on a caloric restriction regimen (-500 kcal/d).

Results: Body weight remained stable for both groups throughout the maintenance study. The high dairy diet resulted in decreases in total body fat (5.4%, $p < 0.01$), trunk fat (4.6, $p < 0.01$), insulin (14%, $p < 0.04$), and blood pressure (6.8 mm Hg systolic, $p < 0.01$; 4.25 mm Hg diastolic, $p < 0.01$) and an increase in lean mass (2.2%, $p < 0.04$), whereas there were no significant changes in the low dairy group. Although both diets produced significant weight and fat loss in the weight loss study, weight and fat loss on the high dairy diet was ~2-fold higher ($p < 0.01$), and loss of lean body mass was markedly reduced ($p < 0.001$) compared with the low dairy diet.

Discussion: Substitution of calcium-rich foods in isocaloric diets reduced adiposity and improved metabolic profiles in obese African Americans without energy restriction or weight loss and augmented weight and fat loss secondary to energy restriction.

Key words: dietary calcium, energy restriction, fat loss, intracellular calcium, vitamin D

Introduction

This laboratory has recently shown that increases in dietary calcium significantly augment weight and fat loss secondary to energy restriction (i.e., a deficit of 500 kcal/d) (1). Data suggest that increases from ~400 to 1200 mg of dietary calcium/d increase weight and fat loss by 26% and 28%, respectively. The use of dairy foods in place of calcium supplements has led to respective increases in weight and fat loss of 70% and 64% (1). These outcomes indicate an "antiobesity" effect from dietary calcium and dairy products, with more pronounced effects from the latter (2). These findings are consistent with those from clinical (3-5), epidemiologic (5-9), and rodent (2) studies.

A compelling set of mechanisms supporting these clinical and epidemiologic observations is provided by our studies of the role of intracellular Ca^{2+} signaling and calcitrophic hormones in regulating human adipocyte metabolism (2). The increase in calcitriol (1,25-dihydroxyvitamin D) elicited by low calcium diets acts on adipocytes through a specific membrane vitamin D receptor to increase Ca^{2+} influx. The resultant rise in intracellular Ca^{2+} exerts a coordinated effect on adipocyte metabolism, serving to stimulate lipogenic gene expression and lipogenesis while simultaneously suppressing lipolysis, thereby increasing lipid filling and adiposity (2,5). In addition, calcitriol acts through a nuclear vitamin D receptor to suppress the expression of adipocyte uncoupling protein 2 expression, thereby limiting mitochondrial fatty acid transport and oxidation (2,10), as well as inhibiting adipocyte apoptosis (11).

Received for review November 22, 2004.

Accepted in final form May 5, 2005.

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Department of Nutrition, The University of Tennessee, Knoxville, Tennessee.

Address correspondence to Michael B. Zemel, Department of Nutrition, The University of Tennessee, 1215 W. Cumberland Avenue, Room 229, Knoxville, TN 37996-1920.

E-mail: mzemel@utk.edu

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Because high calcium diets suppress calcitriol levels, they result in reduced lipogenesis and increases in lipolysis, uncoupling protein 2 expression, and adipocyte apoptosis (2,10,11), thereby reducing lipid filling and adiposity (12,13). Although this framework provides a mechanism for the effects of calcium in modulating adipocyte metabolism and adiposity, it does not provide an explanation for the substantially greater effects observed with dairy vs. supplemental sources of calcium. We have proposed that this augmented effect may be caused by additional bioactive factors, such as angiotensin converting enzyme inhibitors, as well as the rich concentration of branched chain amino acids found in dairy that may act in concert with the calcium to attenuate adiposity (2); however, this concept has not yet been directly shown.

Our previous clinical trial used an energy-restricted diet in a primarily white study population (1). However, because African Americans are at greater risk for obesity and consume significantly less calcium and dairy than the white population, this study was designed to determine the effects of dairy-rich diets on body weight and body fat in obese African-American adults studied under maintenance (eucloric) and energy-restricted conditions.

Research Methods and Procedures

Study Design

This study was conducted in two phases. Phase 1 (maintenance) was designed to determine whether inclusion of low-fat dairy products into the diets of obese African-American adults would result in reductions in body fat with preservation or increase of lean body mass in the absence of caloric restriction. Phase 2 (weight loss) was designed to determine whether a dairy-rich diet would accelerate the fat loss induced by energy restriction in obese African-American adults.

Phase 1 was a 26-week study of 34 otherwise healthy obese African-American adults. Baseline diets were assessed during screening to determine suitability for entry into the study, as described below. Subjects underwent a 2-week lead-in phase to establish a stable baseline of dietary and physiological measures, followed by a 24-week intervention period. At the end of the lead-in period, subjects were randomized to control (low dairy) and high dairy treatment groups. Both groups were given diets isocaloric to those consumed during the lead-in period, with macronutrient and fiber levels maintained equivalent at levels approximating the current U.S. average. The dairy group was given a diet that includes three servings of dairy products per day, with at least one in the form of fluid milk.

Phase 2 followed a similar protocol to that described above for phase 1, but studied 29 otherwise healthy African-American adults under hypocaloric conditions. Subjects were studied for a 2-week lead-in period to establish their

current caloric requirements and provide an opportunity for baseline dietary and physiological assessment, and they were randomized to the following two outpatient dietary regimens for 24 weeks: 1) a control diet providing a 500-kcal/d deficit, zero to one servings of low-fat dairy products per day, and containing a total of ~500 mg calcium per day or 2) a high dairy diet providing a 500-kcal/d deficit and containing three servings of dairy products per day.

Subjects in both phases were provided individual instruction, counseling, and assessment from the study dietitian regarding dietary adherence and the development and reinforcement of strategies for continued success, and diets were monitored weekly.

Body weight and waist circumference were measured weekly, with subjects wearing street clothes with no shoes, outerwear, or accessories. Body fat was measured at the beginning of the study and at weeks 12 and 24 using DXA. DXA was also used to measure changes in regional fat distribution (abdominal vs. other regions). Blood pressure and circulating insulin, glucose, and lipids [triglycerides and total and high-density lipoprotein (HDL)-cholesterol] were measured in the fasting state at the same intervals (baseline and weeks 12 and 24).

Subjects

Thirty-nine otherwise healthy obese African-American adults ranging in age from 26 to 55 years were initially enrolled in the maintenance study, and 36 were initially enrolled in the weight loss study. Of these, 34 completed the maintenance study, and 29 completed the weight loss study. Those who did not complete the study did not exhibit significant differences in any of the baseline characteristics in comparison with those who did complete the study. Reasons for drop-out included scheduling conflicts ($n = 3$ for maintenance; $n = 4$ for weight loss), dissatisfaction with lack of weight loss in the maintenance study ($n = 2$), and reluctance to comply with caloric restriction in the weight loss study ($n = 3$). All subjects had an initial BMI of 30 to 40 kg/m²; a low calcium (<600 mg/d) and low dairy (<1 serving/d) diet, as determined by food frequency and diet history at study entry; no more than a 3-kg weight change over the preceding 12 weeks; and no recent (4 weeks) changes in exercise intensity or frequency. Subjects were excluded from participation if they required the use of oral antidiabetic agents or insulin; used obesity pharmacotherapeutic agents and/or herbal or other preparations intended for use in obesity or weight management; had a history of significant endocrine, hepatic, or renal disease; were pregnant or lactating; or suffered any form of malabsorption syndrome. Seven of the subjects in the weight loss study were on stable antihypertensive pharmacotherapy (high dairy, $n = 4$; low dairy, $n = 3$), which was continued during the study. Subject characteristics are summarized in (Tables 1 and 2).

Table 1. Phase 1 (maintenance) baseline patient characteristics

	Low dairy	High dairy
Sex	9 F, 8 M	14 F, 3 M
Age (years)	41.3 ± 2.7	42.5 ± 2.6
BMI (kg/m ²)	34.9 ± 0.8	34.1 ± 0.7
Systolic blood pressure (mm Hg)	133 ± 3	131 ± 3
Diastolic blood pressure (mm Hg)	84 ± 2	90 ± 2
Total cholesterol (mg/dL)	179 ± 14	243 ± 45
HDL cholesterol (mg/dL)	43 ± 5	47 ± 4
Triglycerides (mg/dL)	165 ± 23	171 ± 31

Values are mean ± SE.

This research was approved by the Institutional Review Board of the University of Tennessee; informed consent was obtained from all subjects, and the research was conducted in accordance with the ethical standards outlined in the Helsinki Declaration.

Diets

Baseline dietary assessments (diet records) were conducted by the project dietitian during the 2-week lead-in period and were used to provide an initial estimate of a maintenance level of caloric intake. This figure was refined by calculating energy needs using World Health Organization equations for calculation of basal metabolic rate, which were adjusted for activity level to provide an estimate of total daily energy expenditure (TDEE)¹. TDEE was calculated as $1.3 \times$ basal metabolic rate for obese patients engaged in mild daily activity and $1.5 \times$ basal metabolic rate for those engaged in strenuous daily activity. Discrepancies between estimated TDEE and baseline caloric intake were resolved, if necessary, by repeat diet records reviewed by the project dietitian. Based on this initial estimate of caloric needs, diets were prescribed that were either eu-caloric (phase 1) or produced a caloric deficit of ~500 kcal/d (phase 2). The diets for the treatment arms were constructed to provide comparable levels of macronutrient and fiber, to approximate the average consumption in the United States (fat, ~35% of total kilocalories, carbohydrates ~49%, protein ~16%, fiber 8 to 12 g/d). The increased dairy consumption in the high dairy group was accomplished primarily by substitution of dairy products for lean meats.

¹ Nonstandard abbreviations: TDEE, total daily energy expenditure; HDL, high-density lipoprotein; LDL, low-density lipoprotein; 11 β -HSD-1, 11 β -hydroxysteroid dehydrogenase-1.

Table 2. Phase 2 (weight loss) baseline patient characteristics

	Low dairy	High dairy
Sex	11 F, 1 M	14 F, 3 M
Age (years)	41.7 ± 2.7	41.7 ± 2.9
BMI (kg/m ²)	35.4 ± 0.9	35.6 ± 0.7
Systolic blood pressure (mm Hg)	126 ± 3	126 ± 3
Diastolic blood pressure (mm Hg)	85 ± 3	84 ± 1
Total cholesterol (mg/dL)	161 ± 8	156 ± 11
HDL cholesterol (mg/dL)	34 ± 2	36 ± 3
Triglycerides (mg/dL)	76 ± 8	71 ± 11

Values are mean ± SE.

Nutritional supplements were not permitted, and caffeine intake was maintained at a constant level (individualized for each patient, based on baseline assessment). Diets were prescribed and monitored as noted above, and key characteristics of the diets achieved by subjects are shown in Table 3.

Assessment

Body weight was measured with a calibrated scale, and height was measured with a wall-mounted stadiometer with subjects in street clothes with no outerwear or shoes. BMI was calculated using standard equation (kilograms per meters squared). Waist circumference was measured in the standing position, with measurements obtained midway between the lateral lower rib margin and the iliac crest. The measurements were taken midexhalation, and the average of two readings was recorded.

Total fat mass and percentage lean and fat mass were assessed by DXA (Lunar Prodigy; Lunar, Madison, WI) at baseline and at weeks 12 and 24 of study. The instrument was calibrated, and a phantom was assessed daily to determine if drift had occurred; spine phantom variation was <2.1% throughout the study.

Blood pressure was measured with subjects seated in an upright position in a chair for at least 5 minutes with the arm supported at heart level. Blood pressure was measured with an appropriately sized cuff using a standard, calibrated sphygmomanometer on the same arm for every measurement. Two readings, at least 1 minute apart, were taken, and the average value was reported.

Plasma glucose was determined using a glucose oxidase method and insulin and leptin levels were determined using standard radioimmunoassay using commercially available kits (Linco Research, St. Charles, MO). Fasting lipid profiles [cholesterol, low-density lipoprotein (LDL)-choles-

Table 3. Diet characteristics

	Maintenance phase		Weight loss	
	Low dairy	High dairy	Low dairy	High dairy
Calcium intake (mg/d)	458 ± 96	1124 ± 53	468 ± 23	1037 ± 27
Energy intake (kcal/d)	1843 ± 98	1982 ± 124	1278 ± 84	1491 ± 62
Percent energy from fat	33 ± 4	34 ± 3	30 ± 2	31 ± 2
Percent energy from protein	17 ± 2	17 ± 1	17 ± 1	18 ± 2
Percent energy from carbohydrate	50 ± 3	49 ± 5	53 ± 3	51 ± 2

Values are mean ± SE.

terol, HDL-cholesterol, and triglycerides) were assessed by standard autoanalyzer techniques in the clinical laboratory.

Statistical Analysis

Data for each study phase was assessed with MANOVA using SAS-PC software to facilitate evaluation of both the repeated-measures and independent group comparisons inherent to this study design. Only subjects who completed the entire study they were enrolled in (weight maintenance or weight loss) were included in the data analysis. All data are presented as mean ± SE.

Results

Phase 1 (Weight Maintenance)

There were no significant changes in body weight over the 24-week study (total weight change of 0.2 ± 0.5 and 0.4 ± 0.6 kg in the low and high dairy groups, respectively). However, there were significant decreases in both total body fat and trunk fat (Figure 1) in the high dairy group, with a corresponding increase in lean body mass (Figure 1). The decrease in total body fat was reflected in a corresponding decrease in circulating leptin, which decreased by 18.7 ± 5.4 ng/mL in the high dairy group ($p < 0.05$) compared with a non-significant increase of 4.2 ± 3.7 ng/mL in the low dairy group. Consistent with the observed changes in trunk fat, there was a significant decrease in waist circumference in the high dairy group, whereas there was no significant change in the low dairy group (Figure 2).

Subjects on the high dairy diet exhibited a significant increase in circulating glycerol ($26.5 \pm 11.1\%$, $p < 0.01$), indicating an increase in lipolysis in individuals on this diet, whereas there was no significant change observed in the low dairy group ($3.6 \pm 7.5\%$). There were significant decreases in both systolic and diastolic blood pressure on the high dairy diet ($p < 0.01$; Figure 3), whereas there were no significant changes in blood pressure in subjects consuming the low dairy diet. Similarly, there was a marked decrease in circulating insulin levels only in subjects consuming the

high dairy diet ($p < 0.05$; Figure 4). Diet was without significant effect on circulating lipids in either dietary treatment group.

Phase 2 (Weight Loss)

The daily energy deficit of 500 kcal/d produced weight and fat loss in all subjects; however, both weight and fat loss were nearly 2-fold greater in subjects consuming the high dairy diet compared with those consuming the low dairy diet ($p < 0.01$; Figure 5). Moreover, although there was a

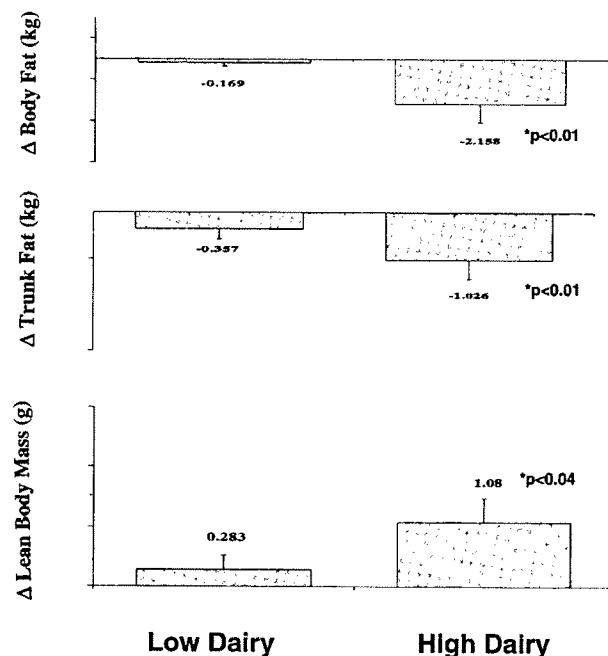


Figure 1: Effects of dietary treatments on total body fat (top), trunk fat (middle), and lean body mass (bottom) in the maintenance study.

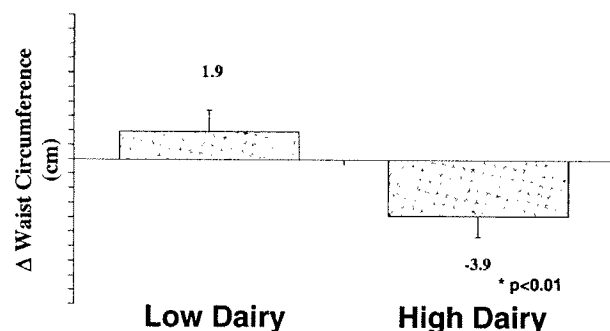


Figure 2: Effects of dietary treatments on waist circumference in the maintenance study.

significant loss of lean body mass in subjects consuming the low dairy diet during energy restriction, loss of lean body mass was markedly attenuated in those consuming the high dairy diet ($p < 0.001$; Figure 6). Trunk fat loss was substantially increased in subjects consuming the high dairy diet vs. the low dairy diet ($p < 0.01$; Figure 7), and this was reflected in correspondingly greater decreases in waist circumference in the high dairy group ($p < 0.05$, Figure 7).

Although circulating glycerol was increased significantly in both groups, there was a significantly greater increase in the high dairy group ($28.0 \pm 8.3\%$ vs. $12.7 \pm 6.8\%$, $p < 0.01$). There was no significant effect of diet on systolic or diastolic blood pressure; however, unlike the maintenance study, seven of the subjects enrolled in the weight loss study (four in high dairy and three in low dairy) were maintained on stable antihypertensive pharmacotherapy at enrollment, and they maintained these regimens throughout the study. Circulating insulin decreased with weight loss in both

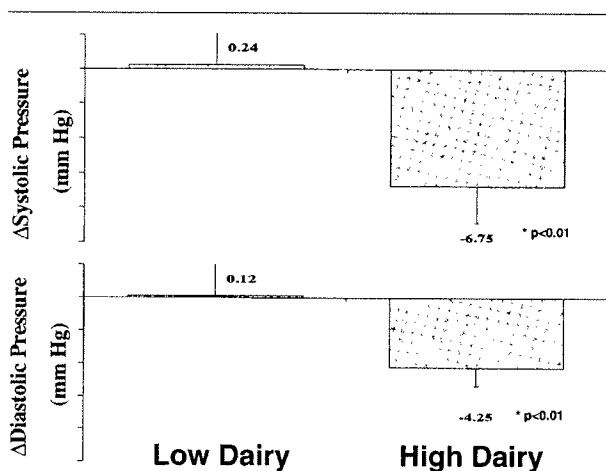


Figure 3: Effects of dietary treatments on systolic pressure (top) and diastolic pressure (bottom) in the maintenance study.

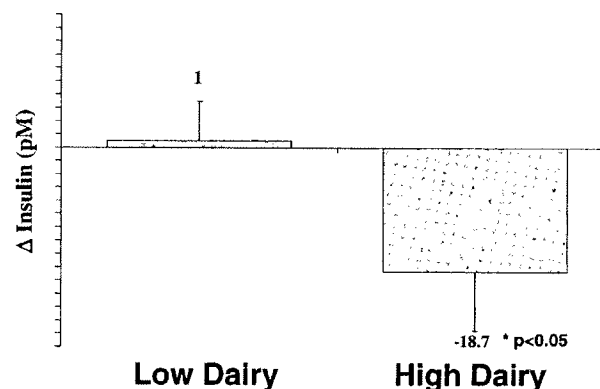


Figure 4: Effects of dietary treatments on circulating insulin in the maintenance study.

groups; however, the decrease was significantly greater in subjects who consumed the high dairy diet ($p < 0.05$; Figure 8).

Discussion

This study suggests that three servings of dairy foods per day produce significant reductions in total and central adiposity in obese African-American adults—an outcome achieved without weight loss or caloric restriction. We also found that dairy foods accelerate loss of weight and total and central adipose tissue mass secondary to energy restriction. The prevalence of overweight and obesity among

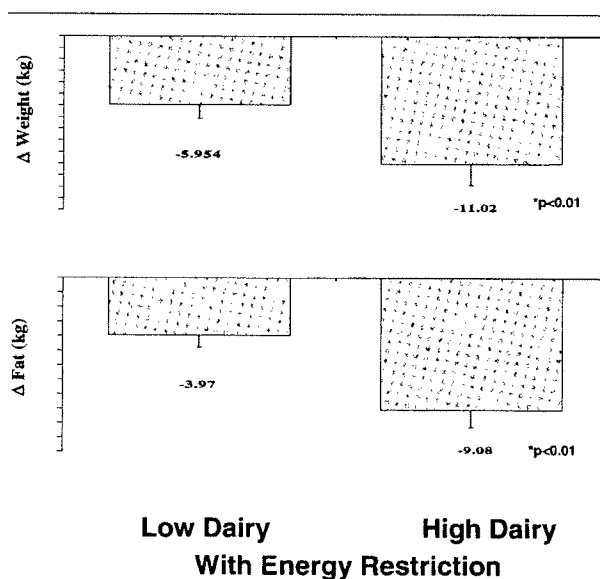


Figure 5: Effects of dietary treatments on weight loss (top) and fat loss (bottom) after weight loss.

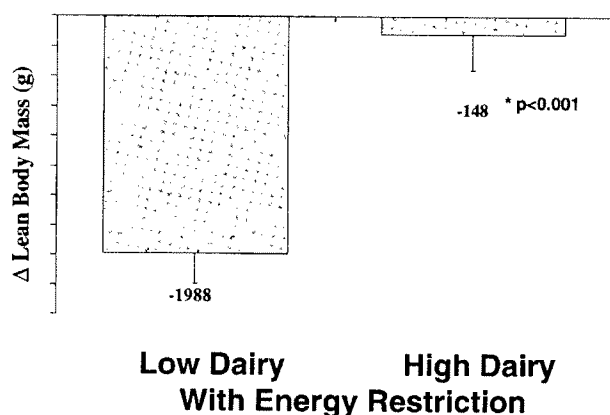


Figure 6: Effects of dietary treatments on lean body mass after weight loss.

African Americans is higher than among U.S. whites (31.1 vs. 19.6% of adults) (14), and 50% of non-Hispanic African-American women are obese (15). Moreover, African Americans are at significantly greater risk than other segments of the U.S. population for consuming suboptimal levels of calcium; <25% of African-American adults consume the recommended daily allowance for calcium, and the average calcium and dairy intakes are 661 mg/d and one dairy serving, respectively, with adult African-American women exhibiting intakes of only 526 mg/d. Our data suggest that suboptimal dairy and calcium intakes may contribute to increased total and central adiposity and obesity in

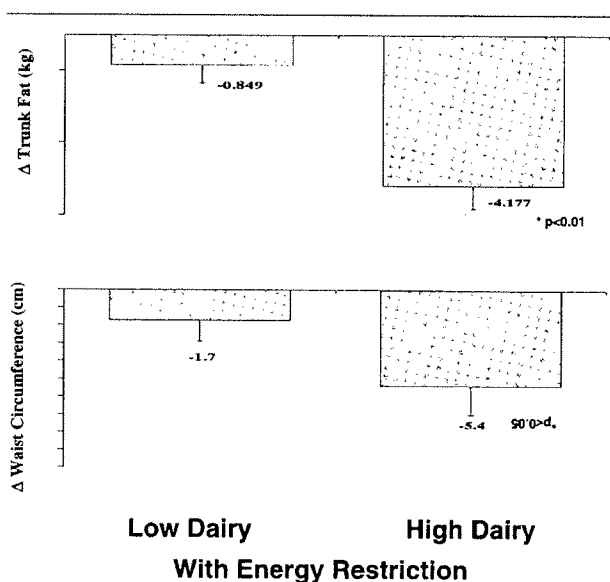


Figure 7: Effects of dietary treatments on trunk fat (top) and waist circumference (bottom) after weight loss.

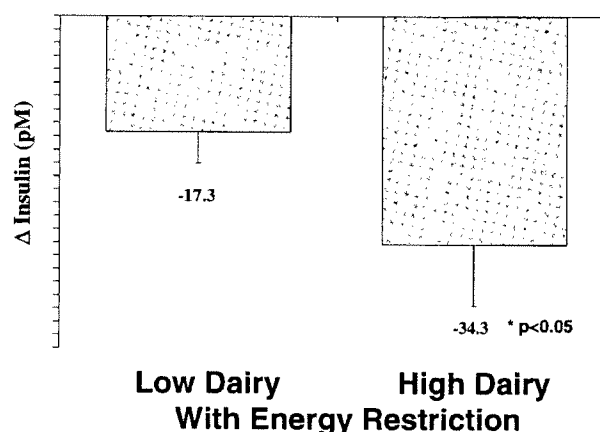


Figure 8: Effects of dietary treatments on circulating insulin after weight loss.

already-obese African-American adults. It may also interfere with successful weight and fat loss during energy restriction. These data are consistent with those from our earlier studies (1,16). Outcomes from the weight maintenance phase of the study are also consistent with prior findings (5).

We found significant increases in circulating glycerol in both the weight loss and maintenance phases of the high dairy diets. These outcomes are consistent with our previous observations of the antilipolytic effects of 1,25-dihydroxyvitamin D in human adipocytes (2,5,17). Similar increases in lipolysis have been seen in animals fed high calcium diets (2,5,12,13). Whole room calorimetry data from Melanson et al. (18,19) are also consistent with these findings.

Data from this study suggest that consumption of a dairy-rich diet confers protection against loss of lean body mass during energy restriction. Similarly, we recently observed partial preservation of lean body mass in whites placed on an energy-restricted diet (16). This outcome may be attributable to the high proportion of branched chain amino acids found in dairy proteins (2). In addition to supporting protein synthesis, the branched chain amino acids, especially leucine, play specific metabolic roles as energy substrates and in the regulation of muscle protein synthesis (20). In skeletal muscle, leucine stimulates protein synthesis and inhibits protein catabolism through multiple independent mechanisms (21–25). Accordingly, only diets that provide leucine at levels in excess of that required for protein synthesis can fully support these additional signaling pathways (20), and the ability of dairy to partially protect skeletal muscle mass during energy restriction may be related to its rich leucine concentration.

We have previously noted preferential loss of central adipose tissue mass in individuals subjected to caloric restriction on high dairy vs. low dairy diets (1,16). Data from

this study extend these observations to African Americans under both weight maintenance and energy-restricted conditions. One possible mechanism in the development of central obesity may involve the role of autocrine production of cortisol in adipose tissue. Human adipose tissue expresses significant 11β -hydroxysteroid dehydrogenase-1 (11β -HSD-1), which generates cortisol from cortisone, and 11β -HSD-1 expression is greater in visceral than in subcutaneous adipose tissue (26,27). Moreover, 11β -HSD-1 is elevated in obese individuals, whereas selective overexpression of 11β -HSD-1 in white adipose tissue of mice results in central obesity (28,29), and homozygous 11β -HSD-1 knockout mice are protected from central obesity (30).

We have recently shown that 1,25-dihydroxyvitamin D stimulates 11β -HSD-1 expression and cortisol production in human adipocytes (31,32). In that high calcium diets suppress 1,25-dihydroxyvitamin D levels, we propose that loss of central adiposity on high dairy diets may be attributable, in part, to suppression of 1,25-dihydroxyvitamin D levels and a consequent reduction in cortisol production by visceral adipocytes.

A growing body of observational and population studies support a role for dietary calcium and dairy foods in controlling excess adiposity. Observational studies suggest an inverse relationship between dietary calcium and/or dairy intake and either body weight or body fat in various populations (33–39). Epidemiological studies support these findings (5,6,7,8,9,40), as do data from Davies et al. (3) and Heaney (4). In contrast, findings from Barr (41) failed to support a relationship between calcium intake and body weight. However, this analysis was based on studies that were not designed or powered to detect weight-related outcomes.

Our prior studies report significant augmentation of weight and fat loss associated with high vs. low dairy diets (1,16). Data from the current trial are consistent with these findings. High dairy diets seem to target loss of central obesity and weight without energy restriction. Data suggest that they also accelerate loss of weight and adiposity while preserving lean body mass during energy restriction. These findings are pertinent in that African Americans are particularly affected by obesity and its related comorbidities.

Acknowledgments

This research was supported by The National Dairy Council.

References

1. Zemel MB, Thompson W, Milstead A, Morris K, Campbell P. Calcium and dairy acceleration of weight and fat loss during energy restriction in obese adults. *Obes Res*. 2004;12:582–90.
2. Zemel MB. Role of calcium and dairy products in energy partitioning and weight management. *Am J Clin Nutr*. 2004;79:907S–12S.
3. Davies KM, Heaney RP, Recker RR, et al. Calcium intake and body weight. *J Clin Endocrinol Metab*. 2000;85:4635–8.
4. Heaney RP. Normalizing calcium intake: projected population effects for body weight. *J Nutr*. 2003;133:268S–70S.
5. Zemel MB, Shi H, Greer B, DiRienzo D, Zemel PC. Regulation of adiposity by dietary calcium. *FASEB J*. 2000;14:1132–8.
6. Jacqmain M, Doucet E, Despres J-P, Bouchard C, Tremblay A. Calcium intake, body composition, and lipoprotein-lipid concentrations in adults. *Am J Clin Nutr*. 2003;77:1448–52.
7. Albertson AM, Good CK, Holschuh NM, Eldridge EL. The relationship between dietary calcium intake and body mass index in adult women from three National dietary intake databases. *FASEB J*. 2004;18:6259.
8. Pereira MA, Jacobs DR, Van Horn L, Slattery ML, Kartashov AI, Ludwig DS. Dairy consumption, obesity, and the insulin resistance syndrome in young adults. The CARDIA study. *JAMA*. 2002;287:2081–9.
9. Loos R, Rankinen T, Leon A, et al. Calcium intake is associated with adiposity in black and white men and white women of the HERITAGE family study. *J Nutr*. 2004;134:1772–8.
10. Shi H, Norman AW, Okamura WH, Sen A, Zemel MB. 1α -25-dihydroxyvitamin D₃ inhibits uncoupling protein 2 expression in human adipocytes. *FASEB J*. 2002;16:1808–10.
11. Sun X, Zemel MB. Role of uncoupling protein 2 (UCP2) expression and 1α ,25-dihydroxyvitamin D₃ in modulating adipocyte apoptosis. *FASEB J*. 2004;18:1430–2.
12. Shi H, DiRienzo D, Zemel MB. Effects of dietary calcium on adipocyte lipid metabolism and body weight regulation in energy-restricted aP2-agouti transgenic mice. *FASEB J*. 2001;15:291–3.
13. Sun X, Zemel MB. Calcium and dairy inhibit weight and fat regain during ad libitum consumption following energy restriction in aP2-agouti transgenic mice. *J Nutr*. 2004;134:3054–60.
14. Mokdad AH, Ford ES, Bowman BA, et al. Prevalence of obesity, diabetes, and obesity-related health risk factors, 2001. *JAMA*. 2003;289:76–9.
15. Flegal KM, Carroll MD, Ogden CL, Johnson CL. Prevalence and trends in obesity among US adults, 1999–2000. *JAMA*. 2003;288:1723–7.
16. Zemel MB, Richards JD, Mathis S, et al. Dairy augmentation of total and central fat loss in obese subjects. *Int J Obes Relat Metab Disord*. 2005;29:391–7.
17. Xue B, Greenberg AG, Kraemer FB, Zemel MB. Mechanisms of intracellular calcium inhibition of lipolysis in human adipocytes. *FASEB J*. 2001;15:2527–9.
18. Melanson EL, Shart TA, Schneider J, et al. Relation between calcium intake and fat oxidation in adult humans. *Int J Obes Relat Metab Disord*. 2003;27:196–203.
19. Melanson EL, Ida T, Donahoo WT, Zemel MB, Hill JO. The effects of low- and high-dairy calcium diets on resting energy expenditure and substrate oxidation. *FASEB J*. 2004;18:566.6.
20. Layman DK. The role of leucine in weight loss diets and glucose homeostasis. *J Nutr*. 2003;133:261s–7s.

21. **Kimball SR, Jefferson LS.** Control of protein synthesis by amino acid availability. *Curr Opin Clin Nutr Metab Care.* 2002;5:63–7.
22. **Rieu I, Sornet C, Bayle G, et al.** Leucine-supplemented meal feeding for ten days beneficially affects postprandial muscle protein synthesis in old rats. *J Nutr.* 2003;133:1198–1205.
23. **MacLean DA, Graham TE, Saltin B.** Branched-chain amino acids augment ammonia metabolism while attenuating protein breakdown during exercise. *Am J Physiol.* 1994;267:E1010–22.
24. **Lynch CJ, Halle B, Fujii H, et al.** Potential role of leucine metabolism in the leucine-signaling pathway involving mTOR. *Am J Physiol.* 2003;285:E854–63.
25. **Liu Z, Jahn LA, Long W, Fryburg DA, Wei L, Barrett EJ.** Branched chain amino acids activate messenger ribonucleic acid translation regulatory proteins in human skeletal muscle, and glucocorticoids blunt this action. *J Clin Endocrinol Metab.* 2001;86:2136–43.
26. **Seckl JR, Walker BR.** 11 β -hydroxysteroid dehydrogenase Type 1—a tissue-specific amplifier of glucocorticoid action. *Endocrinology.* 2001;142:1371–6.
27. **Rusk E, Olsson T, Soderberg S, et al.** Tissue-specific dysregulation of cortisol metabolism in human obesity. *J Clin Endocrinol Metab.* 2001;86:1418–21.
28. **Masuzaki H, Paterson J, Shinyama H, et al.** A transgenic model of visceral obesity and the metabolic syndrome. *Science.* 2001;294:2166–70.
29. **Masuzaki H, Yamamoto H, Kenyon CJ, et al.** Transgenic amplification of glucocorticoid action in adipose tissue causes high blood pressure in mice. *J Clin Invest.* 2003;112:83–90.
30. **Kotelevstev Y, Holmes MC, Burchell A, et al.** 11 β -hydroxysteroid dehydrogenase type 1 knockout mice show attenuated glucocorticoid-inducible responses and resist hyperglycemia in obesity or stress. *Proc Natl Acad Sci USA.* 1997;94:14924–9.
31. **Zemel MB, Sobhani T.** Intracellular calcium modulation of cortisol production in human adipocytes. *FASEB J.* 2003;17:A323.
32. **Morris KL, Zemel MB.** 1,25-dihydroxyvitamin D₃ modulation of adipocyte glucocorticoid function. *Obes Res.* 2005;13:670–7.
33. **Tanasescu M, Ferris AM, Himmelgreen DA, Rodriguez N, Perez-Escamilla R.** Biobehavioral factors are associated with obesity in Puerto Rican children. *J Nutr.* 2000;130:1734–42.
34. **Carruth BR, Skinner JD.** The role of dietary calcium and other nutrients in moderating body fat in preschool children. *Int J Obes Relat Metab Disord.* 2001;25:559–66.
35. **Skinner JD, Bounds W, Carruth BR, Ziegler P.** Longitudinal calcium intake is negatively related to children's body fat indexes. *JADA.* 2003;103:1626–31.
36. **Novotny R, Daida YG, Acharya S, Grove JS, Vogt TM.** Dairy intake is associated with lower body fat and soda intake with greater weight in adolescent girls. *J Nutr.* 2004;134:1905–9.
37. **Lin Y-C, Lyle RM, McCabe LD, et al.** Dairy calcium is related to changes in body composition during a two-year exercise intervention in young women. *J Am Coll Nutr.* 2000;19:754–60.
38. **Lovejoy JC, Champagne CM, Smith SR, deJonge L, Xie H.** Ethnic differences in dietary intakes, physical activity, and energy expenditure in middle-aged, premenopausal women: the Healthy Transitions study. *Am J Clin Nutr.* 2001;74:90–5.
39. **Buchowski MS, Semanya J, Johnson AO.** Dietary calcium intake in lactose maldigesting intolerant and tolerant African-American women. *J Am Coll Nutr.* 2002;21:47–54.
40. **McCarron DA.** Calcium and magnesium nutrition in human hypertension. *Ann Intern Med.* 1983;98:800–5.
41. **Barr SI.** Increased dairy product or calcium intake: Is body weight or composition affected in humans? *J Nutr.* 2003;133:245S–8S.